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RESEARCH ARTICLE

Controlled feeding experiments with diets of different abrasiveness reveal slow development of mesowear signal in goats (*Capra aegagrus hircus*)

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ABSTRACT

Dental mesowear is applied as a proxy to determine the general diet of mammalian herbivores based on tooth-cusp shape and occlusal relief. Low, blunt cusps are considered typical of grazers and high, sharp cusps typical of browsers. However, how internal or external abrasives impact mesowear, and the time frame the wear signature takes to develop, still need to be explored. Four different pelleted diets of increasing abrasiveness (lucerne, grass, grass and rice husks, and grass, rice husks and sand) were fed to four groups of a total of 28 adult goats in a controlled feeding experiment over a 6-month period. Tooth morphology was captured by medical CT scans at the beginning and end of the experiment. These scans, as well as the crania obtained post mortem, were scored using the mesowear method. Comparisons between diet groups showed few significant differences after 6 months, irrespective of whether CT scans or the real teeth were scored. Only when assessing the difference in signal between the beginning and the end of the experiment did relevant, significant diet-specific effects emerge. Diets containing lower phytolith content caused a more pronounced change in mesowear towards sharper cusps/higher reliefs, while the feed containing sand did not result in more extreme changes in mesowear when compared with the same feed without sand. Our experiment suggests that the formation of a stable and hence reliable mesowear signal requires more time to develop than 6 months.

KEY WORDS: Dietary signal, Grit, Tooth wear, Controlled food trials, Ruminant

INTRODUCTION

In 2000, Fortelius and Solounias introduced mesowear analysis – a method to rapidly reconstruct paleodiets based on macroscopic attritive and abrasive wear on the molars of ungulate herbivores by observing cheek-tooth occlusal surfaces (Fortelius and Solounias, 2000). As a result, herbivores can quickly be classified as browsers or grazers by observation of their mesowear profile, defined by each tooth's cusp shape (CS) and occlusal relief (OR). The concept of mesowear suggests that tooth-on-tooth wear, or attrition, creates teeth with sharp, pointed cusps and high OR, which is characteristic of browsers, as their diet is hardly abrasive. For grazers, in contrast, the concept suggests that their teeth are mainly worn down by an abrasive diet, in which plant phytoliths and/or external abrasives such as grit and dust grind down the dental material, resulting in lower OR and blunter cusps (Fortelius and Solounias, 2000; Kaiser, 2000).

In the original mesowear scoring system, only the upper second molar was used, and CS was scored as sharp, round or blunt and OR as high or low (Fortelius and Solounias, 2000; Kaiser, 2000). Since then, the system was expanded to include upper and lower molars (Franz-Odendaal and Kaiser, 2003; Kaiser and Fortelius, 2003; Kaiser and Solounias, 2003), as well as a higher number of differentiated scoring states (e.g. Winkler and Kaiser, 2011). However, a more simplified version of the scoring system using a set of gauges has also been introduced (Mihlbachler et al., 2011). Owing to species- or taxon-specific adaptations and exceptions, various modified mesowear scoring systems have been developed for equids (Kaiser and Fortelius, 2003), lagomorphs (Fraser and Theodor, 2010), rhinoceroses (Taylor et al., 2013), marsupials (Butler et al., 2014) and small mammals (Kropacheva et al., 2017; Ulbricht et al., 2015). Mesowear has also been applied to fossil taxonomic lineages such as Chalicotheriidae (Schulz et al., 2007) and notoungulates (Croft and Weinstein, 2008), and in some cases alternative scoring systems using angles and gauges have been used for taxa such as proboscideans (Saarinen et al., 2015) and xenarthrans (Saarinen and Karme, 2017). Owing to the array of varying mesowear techniques, caution should be applied referring to the methodology of the respective mesowear studies, especially when comparing data from different publications.

In extant ungulates, mesowear is most commonly used to reconstruct paleodiets and paleoecology (Croft and Weinstein, 2008), under the assumption that the wear pattern 'generally reflects a substantial portion of the individual's life in ecological time' (Fortelius and Solounias, 2000). Very young individuals as well as senile individuals are generally excluded from mesowear databases, as their extreme signal is not representative of the general population. The macroscopically visible wear pattern is a guide to answer questions about the average diet of a specific species from a particular location. It has also been used to demonstrate tooth wear variability within a species, when different diets are consumed owing to various factors such as climate variables, seasonality, population-specific habitat differences or sexual segregation (Clauss et al., 2007; Kaiser et al., 2009; Kaiser and Schulz, 2006;

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List of symbols and abbreviations

ADIA	acid-detergent insoluble ash
CS	cusp shape
CSA	cusp shape of the anterior cusp
CSP	cusp shape of the posterior cusp
CT	computed tomography
G	grass diet
GR	grass/rice husk diet
GRS	grass/rice husk/sand diet
L	lucerne diet
m1–3	lower molar
M1–3	upper molar
OR	occlusal relief
p2–4	lower premolar
P2–4	upper premolar

Schulz et al., 2007; Taylor et al., 2016, 2014; Winkler and Kaiser, 2011; Yamada, 2012). However, it should be noted that the studies mentioned above only made assumptions about diet composition under the respective circumstances such as 'the wild' or 'captivity', or referred to the literature. Very few studies correlate actual feeding observations with mesowear signals in individuals of the observed populations (Schulz et al., 2013; Wronski and Schulz-Kornas, 2015). Up to now, only a single study (Solounias et al., 2014) has measured mesowear based on a controlled experimental diet, and in this case a new scoring system was introduced, called 'mesowear III' [with the terms 'mesowear I' describing the Kaiser and Solounias (2003) method scoring more than one cusp per tooth, and the term 'mesowear II' referring to the scoring system of Mihlbachler and Solounias (2006)]. For mesowear III, the shape of the inner enamel bands of molars is scored on both their occlusal-mesial and their distal side. Original mesowear scoring was not applied in that experiment, possibly because the relatively short experimental period (up to 40 days) may not have been long enough for a 'mesowear I or II' signal to develop (Fortelius and Solounias, 2000). Correspondingly, Danowitz et al. (2016) claim that 'mesowear III', designated 'inner mesowear', develops faster than the original or 'outer mesowear'.

The mesowear signal changes with age within a species and at different rates across species (Rivals et al., 2007). However, the length of time this signal takes to develop has not been investigated thus far. Most likely, this period is short in lagomorphs and rodents, where a reliable signal can be obtained (Fraser and Theodor, 2010; Ulbricht et al., 2015) even though the cheek-tooth crowns are completely replaced within 1–2 weeks (Müller et al., 2014, 2015).

In order to test whether, and at which rate, a controlled diet can induce mesowear changes, goats were provided with diets of varying abrasiveness for a period of 6 months. We expected individuals fed with a phytolith-poor diet to develop a high–sharp mesowear signal and individuals fed with a phytolith-rich diet to develop a low–round to blunt mesowear signal. An even more drastic effect was anticipated when the same diet was used with added external abrasives. The change in macroscopic tooth shape was recorded using computed-tomographic (CT) imaging and mesowear scoring of CT scans at the beginning and the end of the experimental period.

MATERIALS AND METHODS

Animals

The animal experiments were performed with approval of the Swiss Cantonal Animal Care and Use Committee Zurich (animal experiment license no. 115/2009). Twenty-eight adult female domestic goats [*Capra aegagrus hircus* (Linnaeus 1758)] of mixed breeds and varying ages (Saanen goats, Chamois Colored goats and Toggenburger goats; mean body mass 60±8 kg, estimated age 3–10 years, exact ages unknown), with unknown previous feeding history, were randomly divided into four groups, each consisting of seven individuals, and kept in a stable with an indoor compartment (40 m^2 per group) consisting of a slatted concrete floor, an area covered by industrial carpet, and an outdoor compartment (12 m^2 per group) with a concrete floor. The duration of the feeding experiment ranged from 182 to 198 days for 24 animals and from 107 to 176 days for four other animals that were euthanized before the end of the experiment owing to reasons unrelated to the study (see the original data file in the Dryad deposit for details on individual animals: doi:10.5061/dryad.658433g). At the end of the experimental period the animals were slaughtered and the skulls were prepared by enzymatic maceration at the Center of Natural History, University of Hamburg, where they are permanently curated in the Mammals Collection.

Diets

All animals were fed lucerne hay and lucerne pellets for *ad libitum* consumption for 2 weeks before the first CT scan and the start of the experimental diet feeding period. The experimental diets differed in abrasiveness between the groups, with increasing abrasiveness from lucerne pellets (L), grass pellets (G) to grass pellets with rice husks (GR) and grass pellets with rice husks with an addition of sand (GRS; sand for playgrounds, grain size 0–1 mm, REDSUN garden products B.V., Heijen, Denmark; mean particle size 0.233 mm). These diets were of the same batch as those used in experiments with rabbits (*Oryctolagus cuniculus*) (Müller et al., 2014), guinea pigs (*Cavia porcellus*) (Müller et al., 2015) and *in vitro* with horse teeth (Karme et al., 2016). During the production of the pelleted diets, care was taken to ensure that the proportion of indigestible abrasives in the GRS diet was mimicked in the other diets by a similar proportion of indigestible, non-abrasive filler, to ensure comparable levels of energy and nutrients per amount of pellets (Müller et al., 2014). Grass hay was provided to all groups except for the lucerne group, which was fed lucerne hay. A total of 1500 g of pelleted food and 100 g of hay was provided daily per animal. It should be noted that in this experiment, the diets were designed to mainly comprise pellets, and the provided proportion of hay was therefore lower than the normal forage ration for ruminants. Water was available for *ad libitum* consumption at all times. Samples of all diet items, as well as feces of all animals, were analyzed for acid-detergent insoluble ash (ADIA) as a measure for silica (abrasives) content (Hummel et al., 2011); a nutritional analysis of the pelleted diets is given in Müller et al. (2014). Diet samples represented a composite sample of each hay and pellet type, blended from individual weekly samples; fecal samples of all animals were collected immediately before slaughter, i.e. after the animals had been on their respective diets for 6 months.

Computed tomography

CT images were acquired from a helical multislice Siemens scanner (Siemens Medical Solutions, Erlangen, Germany) housed at University of Zurich Tierspital. The parameters kept constant throughout were: tube voltage at 120 kVp, image matrix of 512×512 pixels, field of view 980×332 pixels, slice thickness of 0.6 mm and B60s convolution kernel. The animals were scanned at the start of the experiment, while they were in groups but not yet on experimental diets. The scans took place under general anesthesia with ketamine 10 mg kg^{-1} (Ketonarkon®, Streuli Pharma AG, Uznach, Switzerland) and xylazine 0.1 mg kg^{-1} (Xylazin Streuli,

Streuli Pharma AG) intramuscularly. Anesthesia was maintained with isoflurane (Attane®, Provet AG, Lyssach, Switzerland) administered in oxygen using a facemask. The first CT scan was used as baseline for the tooth condition. Another CT scan was performed post mortem.

Mesowear

The goats' skulls were scored in two ways: (1) physically, by inspection of the original skull material, with the aid of a magnifying lens (magnification ×12), and (2) virtually, using the rendered 3D surface model of the CT data. Mesowear was scored on all premolars and molars excluding unworn, extremely worn or otherwise damaged specimens (Kaiser et al., 2009) using the mesowear scoring protocol from Taylor et al. (2013, 2016) adapted from Fortelius and Solounias (2000) and Winkler and Kaiser (2011). In this system, tooth CS of the anterior (CSA) and the posterior molar cusp (CSP) can be scored as sharp (CS 4), round–sharp (CS 3), round (CS 2), round–round (CS 1) and blunt (CS 0). OR was scored by observing the proportion between height and width of the anterior and posterior molar tooth cusps. Based on this, the different scores are high–high (OR 4), high (OR 3), high–low (OR 2), low (OR 1) and flat–negative (OR 0). This scoring system is not species-specific and is suitable for application on ruminants. Because it was our aim to develop a functional understanding, we investigated diet effects of the individual mesowear components (CS and OR) separately, and did not report results from a combined score (where CS and OR scores are combined into a single number, e.g. Kaiser et al., 2009; Winkler and Kaiser, 2011) here. However, a corresponding evaluation [where, to conform with the way mesowear was scored in our experiment, the combined score values are reversed from the continuous system as presented in Winkler and Kaiser (2011), with 0 being the bluntest lowest score and 17 being the sharpest and highest score] is provided in the supplementary material, and this 'ordinal score' is also given in the original data deposited in Dryad (doi:10.5061/dryad.658433g).

The CT datasets were converted to DICOM medical imaging format and rendered into 3D surface models using Amira 5.6 (Mercury Computer Systems/3D Viz group, San Diego, CA, USA) as well as Horos v3.0.1 (Horos Project 2015) for additional visualization. To allow for mesowear scoring in Amira, the view mode was set to orthographic mode and a fixed iso-surface threshold was defined such as to achieve the highest bone resolution while avoiding artefacts. Mesowear scoring of the CT images was performed using a dynamic 3D model of the data; in other words, there was no pre-set, fixed on-screen magnification. Apart from the individual scores, the score difference between the skull and final CT scan was calculated, as well as the score difference between the initial and the final CT scans for each cusp/tooth. All mesowear scoring was performed blind to diet groups, and the scoring of the skulls was performed separately from that of the CT scans. All scoring was performed by the same investigator (N.L.A.), and the descriptive scores (e.g. high, blunt, sharp; summarized in the original data in the Dryad deposit: doi:10.5061/dryad.658433g) were transferred into numerical values as described above. The difference in scores taken from skulls and CT scans was not used to adjust scoring, but were part of this investigation.

Statistical analysis

For statistical analysis, data for left and right teeth were entered separately in the dataset. The overall difference in sharpness between anterior and posterior cusps was tested, as well as the differences between mesowear results from the same time point using different methods (virtually on CT scans and physically on skulls). Comparisons within individuals (such as between anterior and posterior cusps, between the initial and final CT scans, and between the CT and the skull scores) were made using Wilcoxon signed-rank tests for paired samples. The distributions of differences between CT and skull scores were assessed by median, mean, skewness and kurtosis. Comparisons between treatment groups were performed using ranked data and general linear models with Tukey's *post hoc* tests; data were ranked every time anew corresponding to the level of analysis (all teeth, all premolars/molars, all upper premolars/molars and all lower premolars/molars, and each individual tooth position) and compared between the four groups. Comparisons of fecal ADIA levels were made using the original data and an ANOVA with Tukey's *post hoc* test. Correlations between scores were assessed by Spearman's ρ. All analyses were performed in SPSS 22.0 (IBM, Armonk, NY, USA), with the significance level set to $P<0.05$. Although most analyses were non-parametric (including the use of ranked data), graphs are based on means and standard deviations to allow visual interpretation of differences.

RESULTS

Dietary silica content

The ADIA content of lucerne and grass hay was very similar to that of the pelleted diets made from each respective material (Fig. 1A). As intended, the diet including rice husks had a higher ADIA concentration and the diet with added sand had a very high ADIA concentration of 92.8 g per kilogram of dry matter. The fecal ADIA levels differed significantly between the groups (ANOVA, $P<0.001$; Fig. 1B), with a significant difference at *post hoc* testing between the GRS and the other groups ($P<0.001$ in all cases) and no overlap in the 95% confidence intervals of any group means.

Mesowear scores from skulls

At the end of the experiment, mesowear scores from skulls and averaged across all molars show the L group as having higher OR scores compared with the other diet groups. The GRS diet group generally had significantly lower CS scores than the L group, with other diet groups in varying positions according to the level of analysis (Fig. 2, Table 1).

Combining the OR and CS scores into a single variable for measurements performed on skulls, CT1, CT2 and the difference between CT1 and CT2 resulted in equal or less differentiation between the diet groups of our experiments, but in no case in a higher level of differentiation (Tables S1–S3, Fig. S1).

Physical and digital scoring

Comparing mesowear scores from post-mortem CT scans and skulls, the percentage of score differences for the OR parameter resembled a normal distribution (albeit with positive kurtosis) around zero, indicating no systematic discrepancy between skull and CT OR scores (Fig. 3). Both CS score differences showed a similar (less kurtotic) distribution shifted (but not skewed) towards the right (Fig. 3). Statistically, no difference was found between the skull and CT scoring for OR (Table 2), and the two measures were significantly correlated ($P<0.001$ for all teeth as well as premolars and molars separately; $\rho=0.44$ for all teeth and premolars and 0.25 for molars). However, CS scores differed significantly in the overall comparison and in each sub-category, as well as for several individual teeth, with cusps being scored as sharper on average in the physical skulls than in the CT scans (Table 2). Nevertheless, for CSA (premolars: $P<0.001$, $\rho=0.28$; molars: $P=0.004$, $\rho=0.18$) and

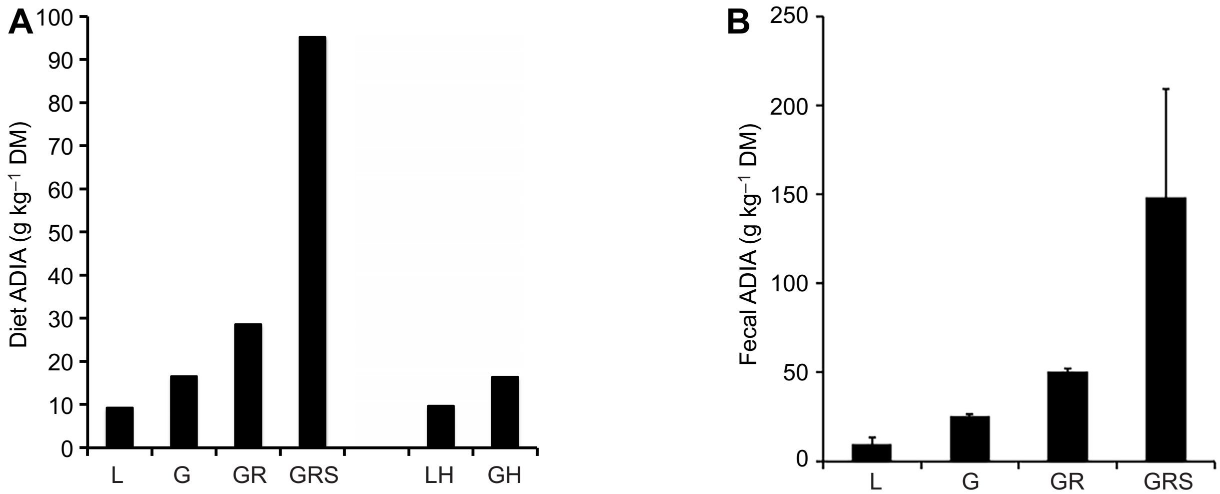

Fig. 1. Silica content of the different diet elements and fecal samples, measured by acid-detergent insoluble ash (ADIA). (A) ADIA content of different pelleted diets and lucerne (LH) and grass hay (GH), supplied for a feeding experiment where goats (*Capra aegagrus hircus*) (n=28) were fed four pelleted diets for 6 months. (B) ADIA (means±s.d.) of fecal samples produced by goats after consumption of different diets. L, lucerne; G, grass; GR, grass/rice husks; GRS, grass/rice husks/sand.

CSP (molars: P=0.005, ρ=0.17), the skull and CT data were significantly correlated as well. Diet did not have a systematic effect on the difference between skull and CT scores (Table 2).

Sharpness of anterior versus posterior cusps

Whether the physical skull or CT scans were scored had an effect on the difference in the perceived sharpness between anterior and posterior cusps. No differences between anterior and posterior cusps in CS scores were found for mesowear scored on the physical skulls (Table S4). For mesowear scored virtually on the CT scans, cusp scores differed significantly in many cases, and in nearly all molars for the second CT scan (Table S4). Diet treatment had no effect on the difference between cusps (Table S4).

Original tooth state

Before the initial CT scan and the beginning of the experimental feeding period, the animals, whose previous diets were unknown, had been kept for 2 weeks on a common diet of lucerne hay and lucerne pellets. Nevertheless, the initial CT scan indicated that significant differences in OR and CSA between the groups had been present before the start of the experiment (with the group to be fed lucerne having higher reliefs and sharper anterior cusps than the other groups in many comparisons), even though animals had been assigned randomly to the groups (Table S5). At the end of the experiment, the results from scoring the final CT scans resembled those of the initial CT scans and those of the skulls, with animals on lucerne having higher relief and sharper cusps than the other groups (Table 3). In pair-wise comparisons, differences between the initial and final CT scores were not significant for OR, but significant differences occurred for CSA for all molars, the lower premolars, the upper M2 and the lower p2, and for CSP in the upper M1 (Table 4). Diet had a significant effect on the difference in mesowear score between the initial and final CT scans, indicating that the L diet had the most distinct (negative) score change, towards higher OR and sharper CS, while the GR diet changed towards lower OR and blunter CS (Table 4; displayed for all molars in Fig. 4 and individual teeth in Fig. 5).

DISCUSSION

The key results of this study show that documenting effects corresponding to current mesowear theory is possible in goats, but that these effects appear to be very small. Our findings further suggest that in ruminants such as goats, mesowear signals may develop at a slower rate than previously assumed.

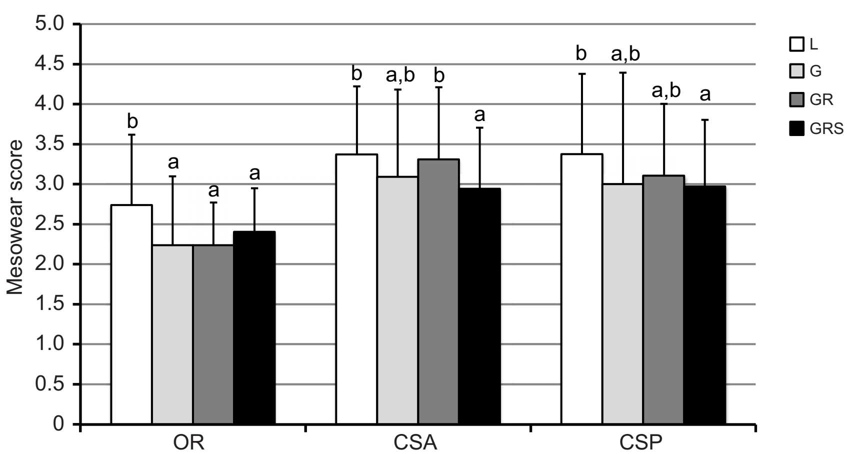

Fig. 2. Averaged mesowear score of all molars, measured on skulls of goats (*Capra aegagrus hircus*) (n=28) fed four pelleted diets of different abrasiveness for 6 months. A higher mesowear score represents higher and sharper teeth. Each diet is represented according to each of the three scoring parameters: occlusal relief (OR), anterior cusp shape (CSA) and posterior cusp shape (CSP). Letters above s.d. error bars represent significance between diets, within score groups. L, lucerne (n=7); G, grass (n=7); GR, grass/rice husks (n=7); GRS, grass/rice husks/sand (n=7).

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Table 1. Effect of different diets on teeth of goats (*Capra aegagrus hircus*) (*n*=28) fed four pelleted diets of different abrasiveness for 6 months, quantified by mesowear occlusal relief and cusp shape of the anterior and posterior cusp on skulls at the end of the experiment

	Occlusal relief skull		Cusp shape anterior skull		Cusp shape posterior skull	
Tooth	*P*	*post hoc*	*P*	*post hoc*	*P*	*post hoc*
All teeth	<0.001	L>G,GR,GRS	<0.001	L>G,GRS; GR>GRS		
All premolars	<0.001	L>G,GR,GRS; GR>GRS	<0.001	L>G,GRS		
All molars	<0.001	L>G,GR,GRS	0.003	L,GR>GRS	0.034	L>GRS
Upper premolars	<0.001	L>G,GRS	0.34	–		
Lower premolars	<0.001	L,GR>GRS	<0.001	L>G,GRS		
Upper molars	0.015	L>GR	0.022	GR>GRS	0.013	L>GR,GRS
Lower molars	0.002	L>G,GR	0.057	–	0.549	–
txP2	0.021	L>GRS	0.351	–		
txP3	0.061	–	0.677	–		
txP4	0.136	–	0.587	–		
txM1	0.443	–	0.025	G<GR	0.199	–
txM2	0.413	–	0.225	–	0.069	–
txM3	0.293	–	0.750	–	0.741	–
tmP2	0.013	L>GRS	0.073	–		
tmP3	0.002	L>G,GRS	0.008	L>G,GRS		
tmP4	0.339	–	0.276	–		
tmM1	0.001	L>G,GR3	0.888	–	0.901	–
tmM2	0.064	–	0.184	–	0.586	–
tmM3	0.531	–	0.143	–	0.788	–

L, lucerne (*n*=7); G, grass (*n*=7); GR, grass/rice husks (*n*=7); GRS, grass/rice husks/sand (*n*=7); tx, maxillary; tm, mandibular. General linear models were performed on ranked data. Tests were performed for all teeth combined, subcategories and individual teeth.

Limitations of study design and method

Upon arrival, the animals' previous diets and exact ages were unknown, and their teeth were in various conditions. Because crown height is one of the parameters that influences mesowear score stability within a species (Rivals et al., 2007), it was important to record tooth state before the beginning of the feeding experiment.

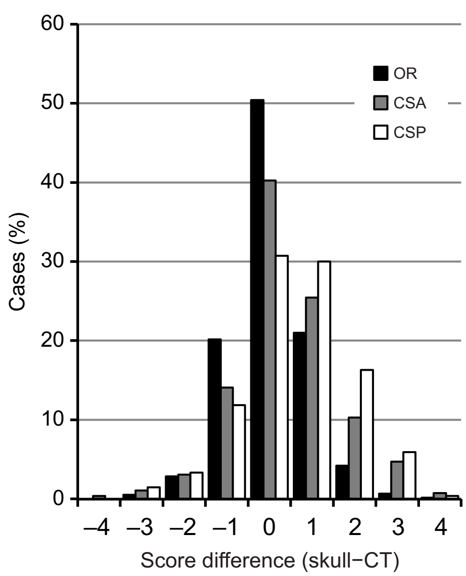

Fig. 3. Difference between the averaged mesowear scores calculated for mesowear variables, taken post mortem on CT images and skulls of goats (*Capra aegagrus hircus*) (*n*=28) fed four pelleted diets of different abrasiveness for 6 months. Distribution of occlusal relief (OR) differences showed a mild positive skew [95% confidence interval (CI) for skewness 0.008 to 0.400] and positive kurtosis (95% CI kurtosis: 0.870 to 1.654) with a median of 0 and a mean of 0.05. Distribution of anterior cusp shape (CSA) differences showed no skew (95% CI: –0.118 to 0.290) and mild positive kurtosis (95% CI: 0.479 to 1.291) with a median of 0 and a mean of 0.38. Distribution of posterior cusp shape (CSP) differences showed no skew (95% CI: –0.464 to 0.116) and no kurtosis (95% CI: –0.397 to 0.759) with a median of 1 and a mean of 0.59.

To dampen this effect, animals could have been sorted into more homogeneous groups based on age and overall tooth wear state after the initial CT scan. Alternatively, the animals could first have been kept for an extensive period of time (given our results, >6 months) on a common diet, though this was beyond our logistical capacities.

In addition, the experimental diets were provided predominantly in pelleted form. Supplying the animals with solely forage-based diets could have been a better representation of the effect of natural forages. However, the logistics of applying external abrasives consistently to this type of diet over long periods of time was considered impractical, and using different natural forages would automatically have introduced other sources of bias, e.g. differences in digestible energy content between forages as well as individual selectivity or inter-group disparity. The pelleted experimental diets used for the present study were specifically designed to be isocaloric and isonitrogenic, in order to avoid differences in the total amount ingested.

Another consideration is the original statement by Fortelius and Solounias (2000) that the ideal sample size for mesowear scoring should be between 10 and 30 individuals. For a preliminary study, such a large number of animals per diet group would have been excessive and unethical. Furthermore, given our results, it appears doubtful that a higher sample size would have demonstrated a larger effect, but only a more significant one.

Finally, combining the OR and CS scores into a single variable did not provide more insight into diet separation. This may be a convenient procedure for further statistical testing of mesowear in relation to other factors, but should not be considered an improvement of the informative value of the score.

Mesowear methodology

According to the original mesowear technique, the sharper of two molar cusps is often used to provide the final tooth score. In the present study, both cusps were scored and these scores were subjected to statistical testing. Thus, the results of our study on goats support the suggestion by Fortelius and Solounias (2000) that there is generally no difference between the sharpness of the anterior and

Table 2. Significance of differences in mesowear scores of teeth of goats (*Capra aegagrus hircus*) (*n*=28) fed four pelleted diets of different abrasiveness for 6 months, from CT scans taken at the end of the experiment and on macerated skulls representing the same time point, as well as the effect of diet on this difference for occlusal relief (OR), anterior cusp shape (CSA) and posterior cusp shape (CSP)

	Difference between skull and CT			Effect of diet on the difference					
	OR	CSA	CSP	OR		CSA		CSP	
Tooth	*P*	*P*	*P*	*P*	*post hoc*	*P*	*post hoc*	*P*	*post hoc*
All teeth	0.257	<0.001	<0.001	0.241	–	0.049	GR>GRS		
All premolars	0.231	<0.001	–	0.068	–	0.214	–		
All molars	0.649	<0.001	<0.001	0.312	–	0.186	–	0.751	–
Upper premolars	0.842	0.001	–	0.039	–	0.781	–		
Lower premolars	0.312	<0.001	–	0.007	GR>GRS	0.044	GR>GRS		
Upper molars	0.144	<0.001	<0.001	0.537	–	0.025	GR>GRS	0.246	–
Lower molars	0.692	0.008	0.030	0.007	L,G<GRS	0.924	–	0.786	–
txP2	0.783	0.425	–	0.958	–	0.647	–		
txP3	0.864	0.035	–	0.411	–	0.866	–		
txP4	0.847	0.007	–	0.505	–	0.920	–		
txM1	0.810	0.108	0.001	0.494	–	0.211	–	0.721	–
txM2	0.336	0.003	<0.001	0.542	–	0.321	–	0.506	–
txM3	0.258	0.015	<0.001	0.681	–	0.582	–	0.248	–
tmP2	0.864	0.153	–	0.643	–	0.828	–		
tmP3	0.105	<0.001	–	0.701	–	0.504	–		
tmP4	0.449	0.036	–	0.091	–	0.115	–		
tmM1	0.321	0.040	0.060	0.344	–	0.764	–	0.910	–
tmM2	0.621	0.333	0.955	0.056	–	0.856	–	0.394	–
tmM3	0.653	0.102	0.025	0.523	–	0.819	–	0.751	–

Comparisons between skulls and CT scans using Wilcoxon signed-rank tests for paired samples. Effect of diet [L, lucerne (*n*=7); G, grass (*n*=7); GR, grass/rice husks (*n*=7); GRS, grass/rice husks/sand (*n*=7)] was tested using general linear models performed on ranked data. tx, maxillary; tm, mandibular. Tests were performed for all teeth combined, subcategories and individual teeth.

posterior cusps within the same molar, at least for measures performed on the skulls.

Originally, the mesowear technique scored only the upper M2 ‘for simplicity’ (Fortelius and Solounias, 2000). Yet, the second molar could be the ideal tooth to represent the mesowear signal, as the first molar erupts first and endures more wear, while the third molar erupts later on and may not have been fully subjected to wear. Our results indicate that of all individual teeth investigated, the second molar showed changes closest to our expectations at statistical significance (in four out of six individual tests in Fig. 5), even though several other teeth, in particular the first and third molar, followed the same trend, albeit less often significantly so.

When scoring mesowear on CT scans, the results indicate OR scoring as similar between CT scans and real skulls. Cusp sharpness, however, is reduced in CT imagery as medical CT scanners are restricted by resolution. The very sharp points are therefore lost at lower resolution, though general relief is conserved (Fig. 6). When results are ranked, diet differences between

Table 3. Significance of differences in the final scores of teeth of goats (*Capra aegagrus hircus*) (*n*=28) fed four pelleted diets of different abrasiveness for 6 months, quantified by mesowear occlusal relief and cusp shape of the anterior and posterior cusp in CT scans at the end of the experiment (corresponding in time point to the scores on the skulls tested in Table 1)

	Occlusal relief final CT		Cusp shape anterior final CT		Cusp shape posterior final CT	
Tooth	*P*	*post hoc*	*P*	*post hoc*	*P*	*post hoc*
All teeth	<0.001	L>G,GR,GRS	<0.001	L>G,GR,GRS		
All premolars	<0.001	L>G,GR,GRS	0.001	L>G,GR,GRS		
All molars	<0.001	L>G,GR,GRS	0.003	L>G,GR,GRS	0.045	L>GR
Upper premolars	<0.001	L>G,GR,GRS	0.052	–		
Lower premolars	0.001	L>GR,GRS	0.003	L>GR		
Upper molars	0.103	–	0.034	L>GR	0.502	–
Lower molars	<0.001	L>GR,GRS	0.14	–	0.074	–
txP2	0.035	L>GRS	0.078	–		
txP3	0.021	L>G	0.368	–		
txP4	0.118	–	0.867	–		
txM1	0.292	–	0.199	–	0.675	–
txM2	0.706	–	0.396	–	0.488	–
txM3	0.195	–	0.237	–	0.271	–
tmP2	0.019	L>GRS, *L>GR*	0.061	–		
tmP3	0.027	L>GRS	0.039	–		
tmP4	0.306	–	0.094	–		
tmM1	0.103	–	0.132	–	0.268	–
tmM2	0.004	L>GR,GRS	0.445	–	0.327	–
tmM3	0.176	–	0.628	–	0.514	–

L, lucerne (*n*=7); G, grass (*n*=7); GR, grass/rice husks (*n*=7); GRS, grass/rice husks/sand (*n*=7); tx, maxillary; tm, mandibular. General linear models were performed on ranked data. Tests were performed for all teeth combined, subcategories and individual teeth.

Table 4. Significance of differences in mesowear scores of teeth of goats (*Capra aegagrus hircus*) (*n*=28) fed four pelleted diets of different abrasiveness for 6 months, from CT scans taken at the beginning and end of the experiment, as well as the effect of diet on this difference for occlusal relief (OR), anterior cusp shape (CSA) and posterior cusp shape (CSP)

	Difference between initial and final CT			Effect of diet on the difference					
	OR	CSA	CSP	OR		CSA		CSP	
Tooth	*P*	*P*	*P*	*P*	*post hoc*	*P*	*post hoc*	*P*	*post hoc*
All teeth	0.231	0.568	0.651	<0.001	L<G,GR,GRS	0.001	L<GR,GRS		
All premolars	0.864	0.121	–	0.004	L<G	0.268	–		
All molars	0.089	0.030	0.651	<0.001	L,G<GR,GRS	<0.001	L<G,GR,GRSG<GR	<0.001	L<GR,GRS GR<GRS
Upper premolars	0.644	0.508	–	0.006	L<G	0.139	–		
Lower premolars	0.101	0.023	–	0.395	–	0.490	–		
Upper molars	0.830	0.142	0.654	0.116	–	<0.001	L<GR,GRS	0.010	L<GR
Lower molars	0.445	0.439	0.885	<0.001	L,G<GR,GRS	<0.001	L<GR,GRS	0.001	L<GR
txP2	0.119	0.197	–	0.052	L<G	0.044	–		
txP3	0.252	0.891	–	0.242	–	0.187	–		
txP4	0.858	0.987	–	0.513	–	0.233	–		
txM1	0.626	0.839	0.036	0.420	–	0.181	–	0.044	*L<GRS*
txM2	0.322	0.027	0.703	0.583	–	0.008	L<GR	0.010	L<GR
txM3	0.176	0.111	0.073	0.162	–	0.038	L<GR	0.921	–
tmP2	0.182	0.008	–	0.258	–	0.019	L>GR		
tmP3	0.364	0.976	–	0.084	–	0.650	–		
tmP4	0.529	0.920	–	0.467	–	0.848	–		
tmM1	0.386	0.508	0.386	0.117	–	0.167	–	0.013	L<GR
tmM2	1.000	0.955	0.396	0.003	L,G<GR	0.003	L<GR	0.343	–
tmM3	0.527	0.136	0.628	0.052	–	0.342	–	0.328	–

Comparisons between initial and final CT scans were made using Wilcoxon signed-rank tests for paired samples. Effect of diet [L, lucerne (*n*=7); G, grass (*n*=7); GR, grass/rice husks (*n*=7); GRS, grass/rice husks/sand (*n*=7)] was tested using general linear models performed on ranked data. tx, maxillary; tm, mandibular. Tests were performed for all teeth combined, subcategories and individual teeth.

individuals are maintained and mesowear scoring remains accurate. It should be noted that 3D renderings do not provide an exact representation of the physical teeth and should therefore only be used to make comparisons within individuals, or within an experimental setup. However, the possibility of applying mesowear scoring to 3D reconstructions of teeth based on medical CT imagery allows expanding the use of mesowear from physical animal skulls to 3D scans of live, sedated animals, and also to observe signal development over time.

Magnitude of mesowear change

The most important finding to emerge from this study is that after spending 6 months on the experimental diet, it was not possible to sort the animals' skulls into correct diet groups based on the mesowear score alone. Even though OR and CS scores showed some significant differences between groups, these differences were of a surprisingly small magnitude (Fig. 2). It was only after using the differential scores between first and last CT scans that testing whether mesowear development followed the predicted theory became possible.

The current literature is ambiguous on the exact time period a mesowear signal takes to develop, and provides vague estimates at best. To this day, no concrete testing of this duration has been made. Fortelius and Solounias (2000) originally state mesowear as reflecting 'a substantial portion of the individual's life in ecological time', whereas others (Jones and Desantis, 2017; Merceron et al., 2007; Yamada, 2012) describe mesowear as representing an average lifelong diet. Some research has defined mesowear as representing an animal's diet over the last few months or years of its life (Rivals et al., 2007; Ulbricht et al., 2015), with some placing the cap at 1 year (Loffredo and DeSantis, 2014; Louys et al., 2012), or even weeks to years, depending on tooth wear state (Danowitz et al., 2016). According to the present study, mesowear signals resulting from feeding pelleted

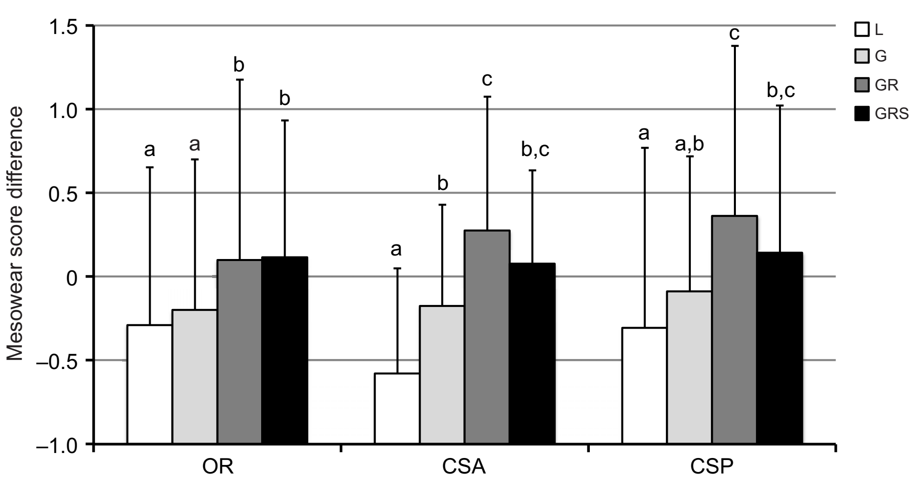

Fig. 4. Averaged score difference between the initial and final CT scan for all molars of goats (*Capra aegagrus hircus*) (*n*=28) fed four pelleted diets of different abrasiveness for 6 months. Each diet is represented according to each of the three scoring parameters: occlusal relief (OR), anterior cusp shape (CSA) and posterior cusp shape (CSP). Letters above s.d. error bars represent significance between diets, within groups. A negative difference indicates that OR became higher, and CS sharper, during the experiment. L, lucerne (*n*=7); G, grass (*n*=7); GR, grass/rice husks (*n*=7); GRS, grass/rice husks/sand (*n*=7).

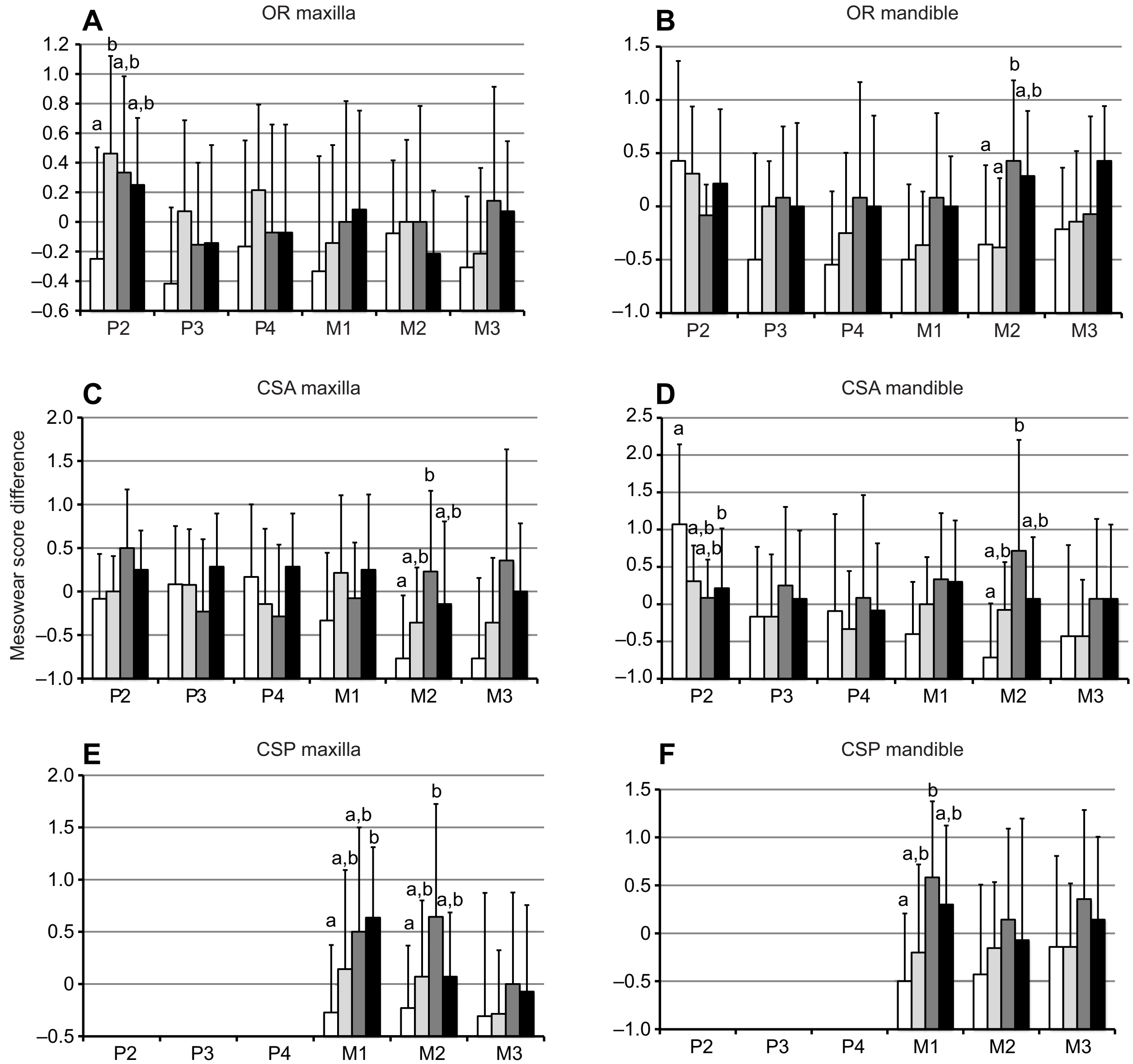

Fig. 5. Averaged score difference between the initial and final CT scan, scored individually for all teeth of goats (*Capra aegagrus hircus*) (*n*=28) fed four pelleted diets of different abrasiveness for 6 months. Letters above s.d. error bars represent significance between diets, within the respective tooth (Table 4). A negative difference indicates that occlusal relief became higher, and cusp shape sharper, during the experiment. (A) Occlusal relief change maxilla. (B) Occlusal relief change mandible. (C) Anterior cusp shape change maxilla. (D) Anterior cusp shape change mandible. (E) Posterior cusp shape change maxilla. (F) Posterior cusp shape change mandible. L, lucerne (*n*=7); G, grass (*n*=7); GR, grass/rice husks (*n*=7); GRS, grass/rice husks/sand (*n*=7).

diets to goats take more than 6 months to develop clearly. Therefore, the assumption that mesowear is of sufficient resolution to track mid-term diet changes such as seasonal diet switches is to be questioned, at least in ruminants. Though most studies tend to use caution when evoking the time frame represented by the mesowear signal, it is often combined with microwear to measure diet seasonality (Kaiser and Schulz, 2006; Mihlbachler and Solounias, 2006; Rivals et al., 2013). Kubo and Yamada (2014) use the standard deviation of the mesowear score for this purpose. If mesowear takes longer than a season to develop, the relationship between mesowear and seasonality will need to be re-evaluated. However, taxon-specific conditions also need be to considered. For example, as mentioned in the Introduction, a mesowear signal in lagomorphs (Fraser and Theodor, 2010; Ulbricht et al., 2015) must develop with 1–2 weeks, given the rate at which cheek-tooth tissue is being replaced.

The change in mesowear score in our study was always less than 1 mesowear unit; more often, this was closer to a change of 0.25. In a very general fashion, one could interpret this as the OR slightly shifting from high towards high–low and the CS shifting from round towards round–round in animals experiencing abrasion. A change of at least 2 mesowear units would be needed to consider animals as having separate diets such as browser or grazer, for example. The small shifts in mesowear signal observed over a period of 6 months brings into consideration the real resolution of the mesowear signal, and offers the possibility of it being more of a lifetime signal in ruminants rather than only representing the last months of an individual's diet. This, in turn, could have an impact on paleontological reconstructions and call for reconsideration of those based on mesowear being a seasonal signal.

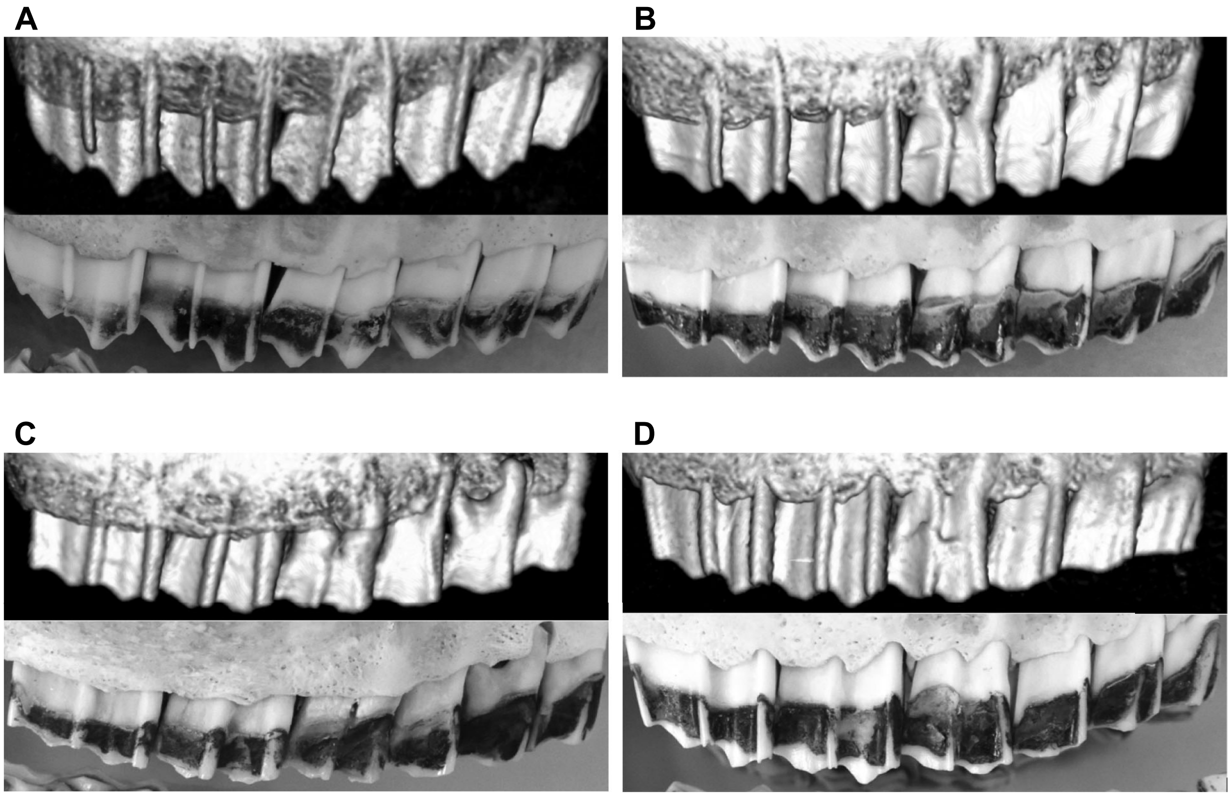

Fig. 6. CT and digital images of the same specimen and position on the upper right tooth row, taken on goats (*Capra aegagrus hircus*) fed four pelleted diets of different abrasiveness for 6 months. (A) Lucerne diet (specimen ID: 13), (B) grass diet (specimen ID: 3), (C) grass/rice husks diet (specimen ID: 27), (D) grass/rice husks/sand diet (specimen ID: 4).

Phytolith effect

The phytolith contents of the experimental diets used in this study are at expected levels based on plant silica content described in Hodson et al. (2005). The hardness of phytoliths and whether they notably affect tooth wear is strongly debated (Baker et al., 1959; Damuth and Janis, 2011; Lucas et al., 2014; Mainland, 2003; Sanson et al., 2007; Xia et al., 2015). In our experimental setting, diets containing more phytoliths, i.e. grass and rice husks versus grass or versus lucerne, caused some abrasive-type wear on ruminant teeth, but the greater change in mesowear (measured as the distance from zero change) was affected by the diet lowest in phytoliths, i.e. lucerne (Fig. 4), suggesting that in our setting, attrition-related sharpening of cusps and deepening valleys was more prominent than abrasion-related blunting of cusps and flattening of valleys. In experiments using the same food with small mammals, tooth wear was not measured by mesowear but by loss of dental tissue via tooth length. In rabbits (Müller et al., 2014) as well as guinea pigs (Müller et al., 2015), when compared within in the same animals (in these animals, repeated measures are logistically possible), the GR diet led to shorter teeth than the G or L diets.

The difference in dietary phytolith content was also hypothesized (though not proven) to be the cause of lower mesowear scores observed in captive giraffe (*Giraffa camelopardalis*) (Clauss et al., 2007), browsing ruminants (Kaiser et al., 2009) and black rhinoceros (*Diceros bicornis*, a browser) (Taylor et al., 2014). These animals putatively receive more grass-related products in captivity than normal for their typically dicot-dominated diets, resulting in abnormally worn teeth. Our observations in the present study concur with the previous assumptions that phytolith-rich diets induce higher rates of dental tissue loss and lead to rounded, lower mesowear profiles, but more prominently emphasize the putative effect of attrition-related change in dental morphology towards sharper teeth.

Grit (sand) effect

The size of external grit as abrasive dietary particles may be an important factor when considering its effect on mesowear. Large-grained sand, similar to that added to the diets in the present study, tends to cause great amounts of wear in cases such as soil ingestion; this has been documented in agricultural (Healy, 1967; Ludwig et al., 1966) and zoo (Jurado et al., 2008) settings. Another illustration of the effect of external abrasives is the case of the pronghorn antelope (*Antilocapra americana*), an antilocaprid seen as an outlier to the known association between grazing and hypsodonty (Janis and Fortelius, 1988). Both hypsodont teeth, usually related to grazing animals and their highly abrasive diet, and a high/sharp mesowear profile, indicating a browser diet, are present in the pronghorn (Damuth and Janis, 2011; Fortelius and Solounias, 2000; Rivals and Semprebon, 2006; Semprebon and Rivals, 2007). As these animals consume only small amounts of grass, one hypothesis for the high hypsodonty and high mesowear score recorded in this species is that their open environment and close-ground feeding behavior results in the ingestion of large amounts of external abrasives in the form of dust (Damuth and Janis, 2011), therefore mandating high-crowned teeth. However, owing to the lack of deviation from the typical browser mesowear signal in the pronghorn, Kaiser et al. (2013) hypothesized that intrinsic abrasives (phytoliths) determine facet development (mesowear appearance), whereas external abrasives in the form of small airborne dust particles could cause uniform loss of dental tissue, shortening the teeth while conserving the mesowear pattern. Experimental studies with external abrasives of a variety of size classes are required to further our understanding of the role of grit and dust.

In the present study, when adding large amounts of coarse abrasives (sand) to the experimental diet, the change in mesowear signal between the start and the end of the experiment was similar to, or lower than, that of the same diet without sand, contrary to our expectations. Additionally, in a controlled microwear study on sheep, where mesowear was not determined, Merceron et al. (2016) found a dietary signal difference in dental microwear texture between monocot and dicot roughage, but no relevant additional signal change when dust was added to these diets. In all likeliness, the key to these differences lays in the way the abrasives (phytoliths versus sand or dust) are embedded in the matrix of the food bolus. Ruminants process food repeatedly through the chewing and regurgitation of cud (Gordon, 1968). Whereas phytoliths are embedded in the plant material, added/exogenous grit may possibly be ‘washed off’ and settle in the rumen, resulting in a less abrasive bolus at regurgitation, which would limit the effects of external abrasives on the teeth (Dittmann et al., 2017; Janis et al., 2010). This ‘washing’ mechanism could explain the lack of effect of sand in goats (present study) and of dust in sheep (Merceron et al., 2016), and provide a reason for why low chewing intensity at ingestion, as compared with rumination, might be adaptive in ruminants (Dittmann et al., 2017). Moreover, the observation in the present study that effects on the sand diet appeared even somewhat lower than the same diet without sand could suggest that the goats chewed the sand-containing diet less intensely at ingestion. In humans, reduced chewing intensity owing to the presence of external abrasives has been documented in a chewing gum study (Prinz, 2004). Whether herbivores, ruminant or not, adjust chewing intensity based on the sensory perception of external abrasives in the ingesta, while appearing probable, remains to be investigated.

This hypothesis further suggests that we expect a distinct difference in the effect of external abrasives on tooth wear between ruminant and non-ruminant herbivores. Indeed, results of the same diets used in (non-ruminant) rabbits and guinea pigs (Müller et al., 2014, 2015) indicate a dramatic effect of the external abrasives. The added-sand diet led not only to the relatively shortest cheek teeth and highest wear (and highest growth rates of the hypseledont cheek teeth), but also to abnormal dental morphology. This reflects the fact that these animals are obliged to chew intensively at ingestion, lacking the option of washing off external abrasives. Additionally, in an *in vitro* study (Karme et al., 2016), again using the same experimental diets, horse teeth were subjected to standardized chewing in a mechanical chewing machine. Actual tooth wear was measured by observing volume loss via micro-CT, and results showed that the GRS diet led to the largest amount of tissue loss, mimicking the results of the same diet in non-ruminants. Overall, these results indicate that a diet containing sand affected the teeth of two non-ruminant species but did not have a comparable effect on the teeth of a ruminant. Although we cannot rule out body size effects, these results highlight the different mechanisms adopted throughout species in order to avoid tooth wear.

Conclusions

The results of this preliminary study infer that in goats, 6 months is not enough time for a mesowear signal to develop conclusively. Accordingly, the lack of a clear distinction between diets after using solely controlled diets for 6 months suggests that in ruminants, the mesowear signal should be considered more as a lifetime or, possibly, depending on future results, an annual rather than a seasonal signal.

The use of CT-based 3D reconstructions for tracing the development of the mesowear signal was an important factor in this study, and further application of this technique could help increase our understanding of mesowear signal development. Similar study designs, including diets with varying sizes and concentrations of grit particles and longer experimental periods, are recommended in order to obtain a solid functional understanding of mesowear-based dietary reconstructions.

Some of the expected effects on mesowear were found, but only once differences between initial and final mesowear scores were observed, a technique difficult to apply in specimens most often subject to mesowear, i.e. fossil or extant-but-deceased specimens. The observed changes indicate, unexpectedly, that diets richer in phytoliths and sand caused less change in mesowear on ruminant teeth than a diet deplete of abrasives (which led to higher/sharper mesowear). Also unexpectedly, the diet with added sand caused mesowear differences similar to those of the same diet without sand, suggesting the possibility that in ruminants, food may be ‘washed’ free of external abrasives when being prepared in the rumen for regurgitation. Thus, the findings also emphasize the relevance of internal abrasives (phytoliths) for the adaptive acquisition of hypsodont molars in ruminants. In the absence of grit effects, internal abrasives may play a more important role in enforcing adaptive responses against tooth wear in ruminants than previously acknowledged.

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Competing interests
The authors declare no competing or financial interests.

Author contributions
Conceptualization: T.M.K., D.W.M., P.R.K., M.C., J.-M.H.; Software: P.R.K.; Formal analysis: N.L.A., M.C.; Investigation: N.L.A., D.W.M., J.H., M.C.; Resources: T.M.K., P.R.K., J.H., J.-M.H.; Data curation: M.C.; Writing - original draft: N.L.A., M.C.; Writing - review & editing: N.L.A., D.E.W., E.S., T.M.K., D.W.M., P.R.K., J.H., M.C., J.-M.H.; Visualization: N.L.A., M.C.; Supervision: E.S., T.M.K., P.R.K., M.C., J.-M.H.; Project administration: M.C., J.-M.H.; Funding acquisition: D.W.M., M.C., J.-M.H.

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Data availability
The original data (all mesowear scores) have been deposited in the Dryad digital repository (Ackermans et al., 2018): https://datadryad.org/resource/doi:10.5061/dryad.658433g/2.

Supplementary information
Supplementary information available online at http://jeb.biologists.org/lookup/doi/10.1242/jeb.186411.supplemental

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Table S1. Effect of different diets on teeth of goats (*Capra aegagrus hircus*) (n=28) fed four pelleted diets of different abrasiveness for six months, quantified by a mesowear score that combines occlusal relief and shape of the anterior cusp in a value between 1-17 (Winkler and Kaiser, 2011) in CTs at the beginning and end, and on the skull at the end of the experiment.

Tooth	CT 1		CT 2		Skull	
	p	*post hoc*	*p*	*post hoc*	*p*	*post hoc*
all teeth	<0.001	L>G,GRS; GR,GRS>G	<0.001	L>G,GR,GRS	<0.001	L>G,GRS; GR>GRS
all premolars	<0.001	L>G,GR,GRS	<0.001	L>G,GR,GRS	<0.001	L>G,GRS; GR>GRS
all molars	<0.001	L,GR,GRS>G	0.002	L>G,GR,GRS	0.007	L>G,GRS
upper premolars	<0.001	L>G,GR,GRS	0.010	L>G,GRS	0.068	-
lower premolars	<0.001	L>G,GR,GRS	0.007	L>GR	<0.001	L>G,GRS
upper molars	0.003	L,GR,GRS>G	0.100	-	0.088	-
lower molars	0.002	L,GR,GRS>G	0.018	L>GR,GRS	0.012	L>G
txP2	0.065	-	0.026	L>GRS	0.165	-
txP3	0.038	L>G	0.203	-	0.193	-
txP4	0.142	-	0.598	-	0.672	-
txM1	0.087	-	0.193	-	0.063	-
txM2	0.244	-	0.370	-	0.413	-
txM3	0.173	-	0.300	-	0.643	-
tmP2	<0.001	L>G,GR,GRS	0.037	L>GRS	0.058	-
tmP3	0.019	L>G,GRS	0.012	L>GR,GRS	0.002	L>G,GRS
tmP4	0.267	-	0.171	-	0.255	-
tmM1	0.013	L>G	0.102	-	0.121	-
tmM2	0.127	-	0.143	-	0.115	-
tmM3	0.278	-	0.505	-	0.119	-

L lucerne (n=7), G grass (n=7), GR grass/rice husks (n=7), GRS grass/rice husks/sand (n=7), tx maxillary, tm mandibular. General Linear Models performed on ranked data. Tests made for all teeth combined, subcategories and individual teeth.

Table S2. Significance of differences of mesowear scores of teeth of goats (*Capra aegagrus hircus*) (n=28) fed four pelleted diets of different abrasiveness for six months, quantified by a mesowear score that combines occlusal relief and shape of the anterior cusp in a value between 1-17 (Winkler and Kaiser, 2011), from CT scans taken at the end of the experiment and on macerated skulls representing the same time point, as well as the effect of diet on this difference.

Tooth	Difference between skull and CT	Effect of diet on the difference	
	p	*p*	*post hoc*
all teeth	<0.001	0.135	-
all premolars	<0.001	0.284	-
all molars	<0.001	0.453	-
upper premolars	0.002	0.909	-
lower premolars	<0.001	0.144	-
upper molars	0.001	0.150	-
lower molars	0.064	0.262	-
txP2	0.305	0.704	-
txP3	0.059	0.995	-
txP4	0.011	0.657	-
txM1	0.223	0.341	-
txM2	0.010	0.399	-
txM3	0.009	0.638	-
tmP2	0.207	0.847	-
tmP3	0.001	0.541	-
tmP4	0.123	0.146	-
tmM1	0.218	0.458	-
tmM2	0.484	0.600	-
tmM3	0.164	0.802	-

Comparisons between skulls and CTs by Wilcoxon signed-rank tests for paired samples. Effect of diet (L lucerne (n=7), G grass (n=7), GR grass/rice husks (n=7), GRS grass/rice husks/sand (n=7)) tested by General Linear Models performed on ranked data. tx maxillary; tm mandibular. Tests made for all teeth combined, subcategories and individual teeth.

Table S3. Significance of differences of mesowear scores of teeth of teeth of goats (*Capra aegagrus hircus*) (n=28) fed four pelleted diets of different abrasiveness for six months, quantified by a mesowear score that combines occlusal relief and shape of the anterior cusp in a value between 1-17 (Winkler and Kaiser, 2011) from CT scans taken at the beginning and end of the experiment, as well as the effect of diet on this difference.

	Difference between initial and final CT	Effect of diet on the difference	
	p	*p*	*post hoc*
all teeth	0.515	<0.001	L,GR,GRS>G
all premolars	0.285	0.046	L>G
all molars	0.056	<0.001	GR,GRS>L,G
upper premolars	0.460	0.143	-
lower premolars	0.100	0.113	-
upper molars	0.396	0.042	GR>G
lower molars	0.278	<0.001	GRS>L,G; GR>G
txP2	0.129	0.571	-
txP3	0.543	0.829	-
txP4	0.719	0.201	-
txM1	0.693	0.669	-
txM2	0.095	0.480	-
txM3	0.169	0.059	-
tmP2	0.012	0.382	-
tmP3	0.687	0.824	-
tmP4	0.722	0.121	-
tmM1	0.750	0.104	-
tmM2	0.989	0.012	GR>L,G
tmM3	0.145	0.018	GRS>G

Comparisons between initial and final CTs by Wilcoxon signed-rank tests for paired samples. Effect of diet (L lucerne (n=7), G grass (n=7), GR grass/rice husks (n=7), GRS grass/rice husks/sand (n=7)) tested by General Linear Models performed on ranked data. tx maxillary; tm mandibular. Tests made for all teeth combined, subcategories and individual teeth.

Table S4. Significance of differences of cusp shape scores for anterior and posterior cusps of the molars of goats (*Capra aegagrus hircus*) (n=28) fed four pelleted diets of different abrasiveness for six months, from CT scans taken at the beginning (CT1) or end (CT2) of the experiment and on macerated skulls, as well as the effect of diet on this difference

Tooth	Difference anterior/posterior cusp			Effect of diet on the difference					
	CT1	CT2	Skull	CT1		CT2		Skull	
	p	*p*	*p*	*p*	*post hoc*	*p*	*post hoc*	*p*	*post hoc*
all molars	<0.001	<0.001	0.139	0.061	-	0.688	-	0.607	-
upper molars	<0.001	<0.001	0.306	0.477	-	0.211	-	0.167	-
lower molars	0.058	0.020	0.209	0.120	-	0.749	-	0.361	-
txM1	0.333	0.011	0.720	0.909	-	0.939	-	0.227	-
txM2	0.674	<0.001	0.295	0.554	-	0.912	-	0.736	-
txM3	<0.001	0.001	0.648	0.350	-	0.108	-	0.579	-
tmM1	0.005	0.023	0.637	0.566	-	0.147	-	0.906	-
tmM2	0.578	0.537	0.806	0.144	-	0.881	-	0.283	-
tmM3	0.827	0.011	0.168	0.104	-	0.873	-	0.878	-

Comparisons between cusps by Wilcoxon signed-rank tests for paired samples. Effect of diet (L lucerne (n=7), G grass (n=7), GR grass/rice husks (n=7), GRS grass/rice husks/sand (n=7)) tested by General Linear Models performed on ranked data. tx maxillary; tm mandibular. Tests made for all teeth combined, subcategories and individual teeth.

Table S5. Significance of differences in the initial scores on teeth of goats (*Capra aegagrus hircus*) (n=28) separated into four groups to be fed four pelleted diets of different abrasiveness for six months, quantified by mesowear occlusal relief and cusp shape of the anterior and posterior cusp on CTs at the beginning of the experiment.

Tooth	**Occlusal relief initial CT**		**Cusp shape anterior initial CT**		**Cusp shape posterior initial CT**	
	p	***post hoc***	***p***	***post hoc***	***p***	***post hoc***
all teeth	<0.001	L>G,GR,GRS	<0.001	L>GR GRS>G		
all premolars	<0.001	L>G,GR,GRS	<0.001	L>G,GR,GRS		
all molars	<0.001	L>G; G<GRS	0.001	L,GR,GRS>G	0.123	-
upper premolars	0.005	L>G,GRS	<0.001	L>G,GR		
lower premolars	0.002	L>G,GRS	<0.001	L>G,GR,GRS		
upper molars	0.029	-	0.13	-	0.452	-
lower molars	0.009	L>G; G<GRS	0.007	G<GR,GRS	0.325	-
txP2	0.066	L>GRS	0.033	*L>G*		
txP3	0.163	-	0.030	L>G		
txP4	0.413	-	0.087	-		
txM1	0.063	-	0.600	-	0.614	-
txM2	0.488	-	0.516	-	0.919	-
txM3	0.242	-	0.358	-	0.641	-
tmP2	0.005	L>GRS	<0.001	L>G,GR,GRS		
tmP3	0.100	-	0.010	L>G		
tmP4	0.450	-	0.105	-		
tmM1	0.040	L>G	0.076	-	0.467	-
tmM2	0.227	-	0.185	-	0.370	-
tmM3	0.478	-	0.249	-	0.990	-

L lucerne (n=7), G grass (n=7), GR grass/rice husks (n=7), GRS grass/rice husks/sand (n=7), tx maxillary, tm mandibular.

General Linear Models performed on ranked data. Tests made for all teeth combined, subcategories and individual teeth.

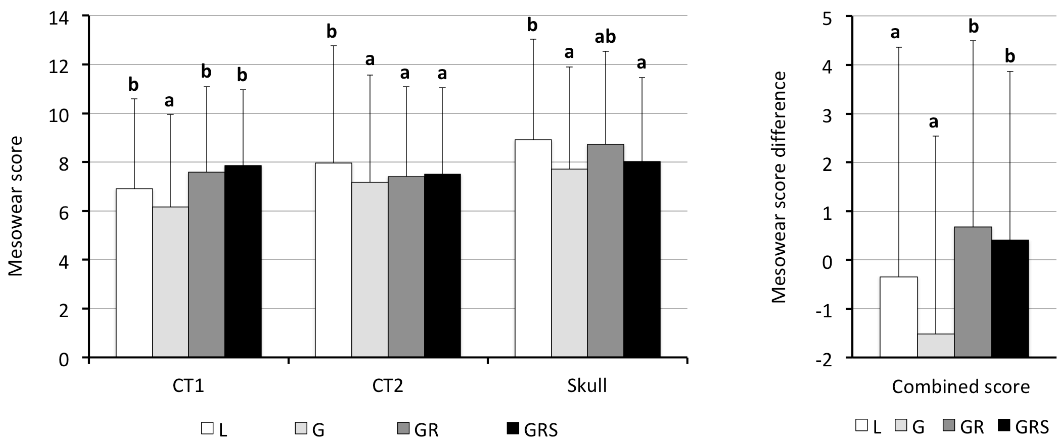

Figure S1. Averaged combined mesowear score, calculated for CT1, CT2 and skulls, as well as the difference in score between CT1 and CT2, for all molars of goats (*Capra aegagrus hircus*) (n=28) fed four pelleted diets of different abrasiveness for six months. A negative difference indicates that OR became higher, and CS sharper, during the experiment. L lucerne (n=7), G grass (n=7), GR grass/rice husks (n=7), GRS grass/rice husks/sand (n=7). Different superscripts indicate significant differences as reported in Table S1 and Table S3.